International Journal on Food, Agriculture, and Natural Resources
Volume 07, Issue 01, Page 1-5
ISSN: 2722-4066
http://www.fanres.org

Original Paper

The Effect of Variations in Baker's Yeast Concentration and Fermentation Time on The Yield and Physicochemical Properties of Virgin Coconut Oil (VCO)

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Received: 31 January 2026; Revised: 27 March 2026; Accepted: 30 March 2026
DOI: *https://doi.org/10.46676/ij-fanres.v7i1.604*

Abstract— **Virgin Coconut Oil (VCO) is a high-value coconut product with superior nutritional quality. This study aimed to evaluate the effect of baker's yeast (Saccharomyces cerevisiae) concentration and fermentation time, as well as their interaction, on the yield and physicochemical properties of VCO. The experiment used a completely randomized design (CRD) with two factors and three replications. The first factor was yeast concentration (0%, 1.5%, and 3% w/v), and the second factor was fermentation time (24, 48, and 72 hours). Parameters observed included yield, moisture content, density, viscosity, and free fatty acid (FFA). Data were analyzed using ANOVA followed by Duncan's Multiple Range Test (DMRT) at a 5% significance level. Results showed that the interaction significantly affected FFA content but had no significant effect on yield and density. Fermentation time significantly influenced moisture content and viscosity, where longer durations (48–72 hours) tended to reduce oil quality. The best treatment was fermentation for 24 hours without yeast addition, which produced comparable yield with better physical quality.**

Keywords—Baker's Yeast, Fermentation, Fermentation Time, Virgin Coconut Oil

I. Introduction

Virgin Coconut Oil (VCO) is an oil product produced from fresh coconut meat without chemical refining or high temperature heating, resulting in a clear liquid with a distinct coconut aroma and taste [1]. Compared to conventional coconut oil, VCO holds higher economic value due to its superior health properties, particularly its high content of Medium Chain Fatty Acids (MCFA) such as lauric acid, which are easily absorbed and metabolized by the body [2]. One of the most potential methods for VCO production, especially for home-scale industries, is fermentation. This method offers several advantages, including a simple procedure, lower production costs, and the preservation of nutritional content due to the absence of thermal processing [3].

The fermentation process relies on the activity of microorganisms to break down the coconut milk emulsion [4]. Baker's yeast, which contains *Saccharomyces cerevisiae*, is commonly used as a starter because it converts natural sugar in coconut milk into ethanol and carbon dioxide, leading to the destabilization of the protein emulsion and the separation of oil [5, 6]. Despite numerous studies on VCO production using fermentation, there is still a lack of comprehensive evaluation that simultaneously examines the interaction between baker's yeast concentration and fermentation time on both yield and detailed physicochemical properties of VCO. Moreover, previous studies often focus on single-factor analysis or limited quality parameters, resulting in fragmented conclusions and the absence of a standardized optimal condition for small-scale or home-based production systems. [4, 7, 8]. Furthermore, the interaction between *S. cerevisiae* activity and environmental factors during fermentation can potentially affect the quality of the oil, such as increasing Free Fatty Acid (FFA) levels due to enzymatic hydrolysis.

The variability in previous findings indicates that the optimal combination of yeast concentration and fermentation time has not been fully established. Inconsistent parameters often lead to product failure or quality degradation, particularly in terms of moisture content and acidity. Despite numerous studies on VCO production using fermentation, there is still a lack of comprehensive evaluation that simultaneously examines the interaction between baker's yeast concentration and fermentation time on both yield and detailed physicochemical properties of VCO. Moreover, previous studies often focus on single-factor analysis or limited quality parameters, resulting in fragmented conclusions and the absence of a standardized optimal condition for small-scale or home-based production systems. Therefore, this study aims to analyze the effect of varying baker's yeast concentrations and fermentation times, as well as their interaction, on the yield and physicochemical properties (moisture content, density, viscosity, and free fatty acid) of VCO to determine the most efficient and optimal treatment.

II. Research Methods

A. Preparation of Tools and Materials

The tools and fermentation containers to be used are checked for damage and then cleaned using a cloth moistened with 70% alcohol solution. The old coconuts aged more or less than 12 months, which are characterized by the brown color of the husk, are peeled and then the flesh is being separated from the shell to be cleaned. The cleaned coconut flesh is then grated [7]. Baker's yeast (*S. cerevisiae*) is also prepared by weighing it for each treatment so that it easier to be applied.

B. Samples Preparation and Fermentation

The grated coconut flesh is added to warm water (36-40°C) at a ratio of 1:1, i.e. 1 kilogram of grated coconut is added to 1 liter of warm boiled water. The grated coconut and water are stirred and then squeezed to extract the coconut milk [9]. The coconut milk is collected in a container and covered to prevent contamination, then left to settle for 2 hours until two layers form, namely a cream layer at the top and a skim layer at the bottom. The layer used as the raw material for making VCO is the top layer, which is then measured at 150 mL and placed in a glass jar.

Baker's yeast was added according to each treatment into glass jars with the following details: 0 gr (0% w/v), 2.25 gr (1.5% w/v), and 4.5 gr (3% w/v). The coconut cream and baker's yeast were then mixed until homogeneous and fermented for varying periods of time, namely 24 hours, 48 hours, and 72 hours. The fermentation jars were covered and left at room temperature.

C. Separation of Coconut Oil

The fermentation results will create three layers, namely oil layer at the top, blondo (coconut curd) layer in the middle, and water layer at the bottom. The oil layer was taken and then filtered using filter paper, then the oils were collected in cleaned and labeled bottles [9].

D. Observation Parameters Analysis

The produced VCO were then tested in the laboratory to determine its yield, free fatty acid content, moisture content, density, and viscosity. The following are the methods used to conduct each test.

1) Yield analysis: The yield measurement was calculated gravimetrically by comparing the weight of the produced VCO with the weight of the coconut cream used. The calculation followed the formula described by Setyorini and Lusiani [10]:

$$\text{Yield (\%)} = \frac{\text{Weight of VCO (grams)}}{\text{Weight of coconut cream (grams)}} \times 100\%$$

2) Free Fatty Acid (FFA) analysis: Based on [11], FFA content were determined using the alkalimetric titration method. A 5 gr VCO sample was dissolved in 50 mL of 97% neutral ethanol and heated. Phenolphtalein (PP) indicator (3-5 drops) was added, and the solution was titrated with standardized 0.1 N NaOH until a stable pink color persisted for 15 seconds. The FFA percentage was calculated as lauric acid equivalent.

$$\text{FFA (\%)} = \frac{\text{V NaOH} \times \text{N NaOH} \times \text{MW VCO}}{1000 \times \text{Sample weight}} \times 100$$

Note:
V NaOH = Volume of NaOH used for titration N NaOH
N NaOH = Normality of NaOH
MW VCO = Molecular Weight of lauric acid (200)

3) Moisture content analysis: Moisture content was measured using the ovendrying method (gravimetric). A 5 gr VCO sample was weighed into a pre-dried crucible and heated in an oven at 105°C for 1 hour. The sample was cooled in a desicator and weighed. This process was repeated until a constant weight was achieved. Moisture content was calculated based on the weight loss of the sample [11].

$$\text{Moisture content} = \frac{a - b}{c} \times 100\%$$

Note:
a = Weight of the crucible and initial sample (grams)
b = Weight of the crucible and final sample (grams)
c = Weight of the initial sample (grams)

4) Density analysis: Density was measured using a pycnometer at 25°C. The empty pycnometer, pycnometer filled with distilled water, and pycnometer filled with the VCO sample were weighed sequentially. The density of VCO was calculated by comparing the mass of the VCO to the mass of the distilled water [12].

$$\rho\ \text{VCO} = \frac{\text{mass of VCO}}{\text{mass of distilled water}} \times \rho\ \text{distilled water}$$

Note:
ρ = Density (g/mL)

5) Viscosity analysis: viscosity was determined using an Ostwald viscometer. The VCO sample was poured into the viscometer and the time required for the liquid to flow between two marked points was measured using a stopwatch. The flow time of the sample was compared to that of distilled water to calculate the viscosity [13].

$$\eta_1 = \eta_2 \frac{t_1 \times \rho_1}{t_2 \times \rho_2}$$

Note:
η_1 = Viscosity of VCO
η_2 = Viscosity of distilled water
t_1 = Flow time of VCO in the viscometer
t_2 = Flow time of distilled water in the viscometer
ρ_1 = Density of VCO
ρ_2 = Density of distilled water

III. Results and Discussion

A. Effect of Treatment Interaction on Free Fatty Acid (FFA)

TABLE I. Free Fatty Acid (FFA) content of VCO at various baker's yeast concentrations and fermentation times

Baker's Yeast Concentration	**Fermentation Time**		
	24 hours	***48 hours***	***72 hours***
0%	2.2 (a) A	3.1 (a) A	1.8 (a) A

Baker's Yeast Concentration	Fermentation Time		
	24 hours	*48 hours*	*72 hours*
1.5%	2.6 (a) A	1.8 (a) A	2.9 (a) A
3%	5 (b) B	2.7 (a) A	2.7 (a) A

Note:

- Numbers followed by the same lowercase letter (horizontal) indicate no significant difference in the effect of fermentation time at the same concentration level of baker's yeast.
- Numbers followed by the same capital letter (vertical) indicate no significant difference in the effect of baker's yeast concentration at the same time level of fermentation time.

The Analysis of Variance (ANOVA) revealed that the interaction between baker's yeast concentration and fermentation time had a significant effect ($p < 0.05$) on the Free Fatty Acid (FFA) content of the produced Virgin Coconut Oil (VCO). However, the FFA levels observed in this study ranged from 1.8% to 5.0%, which significantly exceeded the maximum limit of 0.2% established by the Indonesian National Standard (SNI 7381:2022) [14]. This elevated acidity indicates that the fermentation process facilitated by *Saccharomyces cerevisiae* accelerated the hydrolysis of the oil.

The highest FFA content (5.0%) was recorded in the treatment combining a 3% yeast concentration with a 24-hour fermentation period. This finding suggests that a higher inoculum concentration triggered a substantial release of extracellular lipase enzymes by *S. cerevisiae*, subsequently hydrolyzing triglycerides into glycerol and free fatty acids [15]. While *S. cerevisiae* is primarily utilized for its ability to produce alcohol and lower pH to destabilize the coconut milk emulsion, its secondary lipolytic activity proved detrimental to the chemical quality of the oil in this study.

Notably, the data demonstrated a fluctuation in FFA levels as fermentation time increased. For instance, at a 1.5% yeast concentration, FFA levels decreased from 2.6% at 24 hours to 1.8% at 48 hours, before rising again at 72 hours. This phenomenon may be attributed to the dual metabolic pathways of the yeast. In the initial phase, enzymatic hydrolysis is dominant. However, as sugar substrates become depleted, *S. cerevisiae* may shift its metabolism towards the β-oxidation pathway [16], utilizing free fatty acids as an alternative energy source, thereby causing a temporary reduction in FFA levels. In the final phase (72 hours), cell lysis likely releases intracellular enzymes, re-initiating hydrolysis and causing FFA levels to rebound.

The high Free Fatty Acid (FFA) levels observed in all treatments, which exceeded the SNI 7381:2022 standard (>0.2%), can be attributed to several factors related to the fermentation process. One of the primary causes is the prolonged fermentation time, which enhances lipolytic activity and accelerates the hydrolysis of triglycerides into free fatty acids and glycerol. The presence of microorganisms, including *Saccharomyces cerevisiae*, may indirectly contribute to this process by producing enzymes or creating conditions that favor endogenous lipase activity. In addition, cell lysis during extended fermentation (48–72 hours) can release intracellular enzymes such as lipase into the medium, further increasing FFA formation [15,16].

Another contributing factor is the high moisture content retained in the system, which facilitates hydrolytic reactions. Water acts as a reactant in lipid hydrolysis, and insufficient separation of the aqueous phase from the oil phase can promote continuous FFA formation. Moreover, the absence of thermal treatment in fermentation-based methods, while beneficial for preserving nutrients, also limits enzyme inactivation, allowing lipase activity to persist throughout the process. Raw material quality may also influence FFA levels, as the use of coconut with higher initial moisture or slight deterioration can increase susceptibility to hydrolysis [15].

Therefore, the elevated FFA values indicate that fermentation conditions were not optimal in suppressing hydrolytic reactions. This suggests the need for improved process control, such as shortening fermentation time, optimizing moisture removal, or incorporating post-processing techniques like centrifugation or mild heating to reduce enzymatic activity and achieve compliance with national quality standards.

B. *Effect of Fermentation Time on Moisture Content and Viscosity*

Fermentation time was identified as the sole factor significantly affecting ($p < 0.05$) both moisture content and viscosity. The results exhibited a linear trend wherein extending the fermentation duration resulted in increased moisture content and viscosity. Based on Fig. 1 the lowest moisture content was 0.181% and based on Fig. 2 the lowest viscosity was 48.9 cP, both were achieved at the 24-hour fermentation mark. The following are the graphics of fermentation time effect on moisture content and viscosity sequentially:

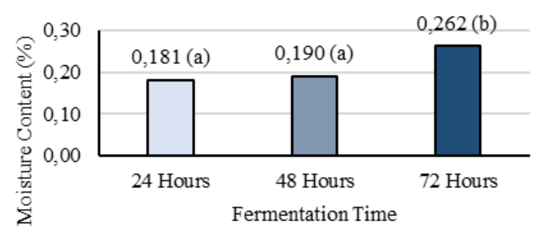

Fig. 1. Effect of fermentation time on moisture content of VCO

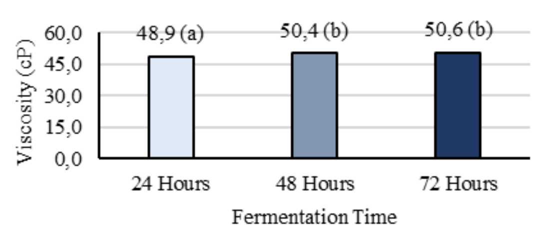

Fig. 2. Effect of fermentation time on viscosity of VCO

Prolonging fermentation to 48 and 72 hours resulted in a significant deterioration of these physical parameters. This decline in quality is attributed to the autolysis of yeast cells and indigenous microorganisms following the stationary phase [8].

Cell lysis releases hydrophilic intracellular components, such as cytoplasmic proteins and cell wall mannoproteins, into the oil phase [7]. These compounds act as natural emulsifiers, binding water molecules and forming micro-emulsions that increase fluid resistance to flow (viscosity) and impede effective water separation during filtration. Consequently, a shorter fermentation duration of 24 hours is recommended to maintain the physical purity of the oil.

C. Yield and Density

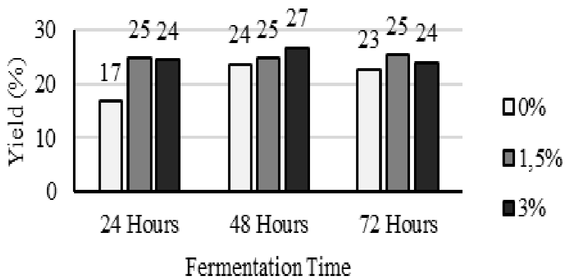

Fig. 3. Effect of baker's yeast concentration and fermentation time on the yield of VCO

Statistical analysis indicated that neither yeast concentration, fermentation time, nor their interaction had a significant effect ($p > 0.05$) on yield and density. Based on Fig. 3, the VCO yield obtained in this study ranged from 17% to 27%. This is lower than the yield reported in a similar study by [9], which achieved up to 35.5%. The lower yield is likely due to the physiological maturity of the raw material; some coconuts used in this study may have not been old enough or have entered the germination phase (haustorium formation), during which oil reserves are catabolized to support shoot growth, thereby reducing recoverable oil content [9].

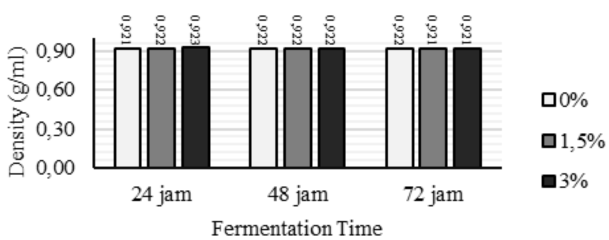

Fig. 4. effect of baker's yeast concentration and fermentation time on the density of VCO

Regarding density, based on Fig. 4. the values obtained (0.921–0.923 g/mL) were slightly above the SNI standard range of 0.915–0.920 g/mL. Correlation analysis revealed a moderately strong positive relationship between moisture content and density ($r = 0.577$). This suggests that the elevated density is primarily caused by residual water [10] and cellular impurities trapped within the oil, which possess a higher density than pure oil. Given that variations in yeast concentration and time did not significantly improve yield, the application of a 24-hour fermentation period without yeast addition (control) is considered the most efficient approach for household-scale production.

IV. CONCLUSION

The interaction between baker's yeast concentration and fermentation time significantly affected the Free Fatty Acid (FFA) content of VCO, but had no significant effect on yield and density. Fermentation time was identified as the main factor influencing moisture content and viscosity, with longer durations (48–72 hours) reducing physical quality. The addition of yeast did not significantly improve oil yield compared to the control treatment. All treatments resulted in FFA levels exceeding the SNI 7381:2022 standard, indicating excessive hydrolysis during fermentation. Therefore, the optimal condition is fermentation for 24 hours without yeast addition (0%), as it provides better efficiency and maintains physical quality.

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